



PCT/US2004/040844

SEQUENCE LISTING

<110> National Institutes of Health

5 Qasba, Pradman
 Boeggeman, Elizabeth
 Ramakrishnan, Boopathy

<120> Catalytic Domains Of Beta(1,4)-Galactosyltransferase I Having
10 Altered Metal Ion Specificity

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10 20 25 30

His Leu Gly Val Thr Leu Val Tyr Tyr Leu Ala Gly Arg Asp Leu Ser

35 40 45

Arg Leu Pro Gln Leu Val Gly Val Ser Thr Pro Leu Gln Gly Gly Ser

50 55 60

15Asn Ser Ala Ala Ala Ile Gly Gln Ser Ser Gly Asp Leu Arg Thr Gly

65 70 75 80

Gly Ala Arg Pro Pro Pro Leu Gly Ala Ser Ser Gln Pro Arg Pro

85 90 95

Gly Gly Asp Ser Ser Pro Val Val Asp Ser Gly Pro Gly Pro Ala Ser

20 100 105 110

Asn Leu Thr Ser Val Pro Val Pro His Thr Thr Ala Leu Ser Leu Pro

115 120 125

Ala Cys Pro Glu Glu Ser Pro Leu Leu Val Gly Pro Met Leu Ile Glu

130 135 140

25Phe Asn Met Pro Val Asp Leu Glu Leu Val Ala Lys Gln Asn Pro Asn

145 150 155 160

Val Lys Met Gly Gly Arg Tyr Ala Pro Arg Asp Cys Val Ser Pro His

165 170 175

Lys Val Ala Ile Ile Ile Pro Phe Arg Asn Arg Gln Glu His Leu Lys

30 180 185 190

Tyr Trp Leu Tyr Tyr Leu His Pro Val Leu Gln Arg Gln Gln Leu Asp

195 200 205

Tyr Gly Ile Tyr Val Ile Asn Gln Ala Gly Asp Thr Ile Phe Asn Arg

210 215 220

35Ala Lys Leu Leu Asn Val Gly Phe Gln Glu Ala Leu Lys Asp Tyr Asp

225 230 235 240

Tyr Thr Cys Phe Val Phe Ser Asp Val Asp Leu Ile Pro Met Asn Asp

245 250 255

His Asn Ala Tyr Arg Cys Phe Ser Gln Pro Arg His Ile Ser Val Ala

40 260 265 270

Met Asp Lys Phe Gly Phe Ser Leu Pro Tyr Val Gln Tyr Phe Gly Gly

275 280 285

Val Ser Ala Ser Ser Lys Gln Gln Phe Leu Thr Ile Asn Gly Phe Pro
290 295 300
Asn Asn Tyr Trp Gly Trp Gly Glu Asp Asp Asp Ile Phe Asn Arg
305 310 315 320
5Leu Val Phe Arg Gly Met Ser Ile Ser Arg Pro Asn Ala Val Val Gly
325 330 335
Thr Cys Arg Met Ile Arg His Ser Arg Asp Lys Lys Asn Glu Pro Asn
340 345 350
Pro Gln Arg Phe Asp Arg Ile Ala His Thr Lys Glu Thr Met Leu Ser
10 355 360 365
Asp Gly Leu Asn Ser Leu Thr Tyr Gln Val Leu Asp Val Gln Arg Tyr
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Pro Leu Tyr Thr Gln Ile Thr Val Asp Ile Gly Thr Pro Ser
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35 40 45
Arg Leu Pro Gln Leu Val Gly Val Ser Ser Thr Leu Gln Gly Gly Thr
50 55 60
30Asn Gly Ala Ala Ala Ser Lys Gln Pro Pro Gly Glu Gln Arg Pro Arg
65 70 75 80
Gly Ala Arg Pro Pro Pro Leu Gly Val Ser Pro Lys Pro Arg Pro
85 90 95
Gly Leu Asp Ser Ser Pro Gly Ala Ala Ser Gly Pro Gly Leu Lys Ser
35 100 105 110
Asn Leu Ser Ser Leu Pro Val Pro Thr Thr Gly Leu Leu Ser Leu
115 120 125
Pro Ala Cys Pro Glu Glu Ser Pro Leu Leu Val Gly Pro Met Leu Ile
130 135 140
40Asp Phe Asn Ile Ala Val Asp Leu Glu Leu Leu Ala Lys Lys Asn Pro
145 150 155 160

Glu Ile Lys Thr Gly Gly Arg Tyr Ser Pro Lys Asp Cys Val Ser Pro
 165 170 175
 His Lys Val Ala Ile Ile Ile Pro Phe Arg Asn Arg Gln Glu His Leu
 180 185 190
 5Lys Tyr Trp Leu Tyr Tyr Leu His Pro Ile Leu Gln Arg Gln Gln Leu
 195 200 205
 Asp Tyr Gly Ile Tyr Val Ile Asn Gln Ala Gly Asp Thr Met Phe Asn
 210 215 220
 Arg Ala Lys Leu Leu Asn Ile Gly Phe Gln Glu Ala Leu Lys Asp Tyr
 10225 230 235 240
 Asp Tyr Asn Cys Phe Val Phe Ser Asp Val Asp Leu Ile Pro Met Asp
 245 250 255
 Asp Arg Asn Ala Tyr Arg Cys Phe Ser Gln Pro Arg His Ile Ser Val
 260 265 270
 15Ala Met Asp Lys Phe Gly Phe Ser Leu Pro Tyr Val Gln Tyr Phe Gly
 275 280 285
 Gly Val Ser Ala Leu Ser Lys Gln Gln Phe Leu Ala Ile Asn Gly Phe
 290 295 300
 Pro Asn Asn Tyr Trp Gly Trp Gly Gly Glu Asp Asp Asp Ile Phe Asn
 20305 310 315 320
 Arg Leu Val His Lys Gly Met Ser Ile Ser Arg Pro Asn Ala Val Val
 325 330 335
 Gly Arg Cys Arg Met Ile Arg His Ser Arg Asp Lys Lys Asn Glu Pro
 340 345 350
 25Asn Pro Gln Arg Phe Asp Arg Ile Ala His Thr Lys Glu Thr Met Arg
 355 360 365
 Phe Asp Gly Leu Asn Ser Leu Thr Tyr Lys Val Leu Asp Val Gln Arg
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 Tyr Pro Leu Tyr Thr Gln Ile Thr Val Asp Ile Gly Thr Pro Arg
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<212> PRT

35<213> Bos taurus

<400> 6

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His Leu Gly Val Thr Leu Val Tyr Tyr Leu Ala Gly Arg Asp Leu Arg
 35 40 45
 Arg Leu Pro Gln Leu Val Gly Val His Pro Pro Leu Gln Gly Ser Ser
 50 55 60
 5 His Gly Ala Ala Ala Ile Gly Gln Pro Ser Gly Glu Leu Arg Leu Arg
 65 70 75 80
 Gly Val Ala Pro Pro Pro Leu Gln Asn Ser Ser Lys Pro Arg Ser
 85 90 95
 Arg Ala Pro Ser Asn Leu Asp Ala Tyr Ser His Pro Gly Pro Gly Pro
 10 100 105 110
 Gly Pro Gly Ser Asn Leu Thr Ser Ala Pro Val Pro Ser Thr Thr Thr
 115 120 125
 Arg Ser Leu Thr Ala Cys Pro Glu Glu Ser Pro Leu Leu Val Gly Pro
 130 135 140
 15 Met Leu Ile Glu Phe Asn Ile Pro Val Asp Leu Lys Leu Ile Glu Gln
 145 150 155 160
 Gln Asn Pro Lys Val Lys Leu Gly Gly Arg Tyr Thr Pro Met Asp Cys
 165 170 175
 Ile Ser Pro His Lys Val Ala Ile Ile Leu Phe Arg Asn Arg Gln
 20 180 185 190
 Glu His Leu Lys Tyr Trp Leu Tyr Tyr Leu His Pro Met Val Gln Arg
 195 200 205
 Gln Gln Leu Asp Tyr Gly Ile Tyr Val Ile Asn Gln Ala Gly Glu Ser
 210 215 220
 25 Met Phe Asn Arg Ala Lys Leu Leu Asn Val Gly Phe Lys Glu Ala Leu
 225 230 235 240
 Lys Asp Tyr Asp Tyr Asn Cys Phe Val Phe Ser Asp Val Asp Leu Ile
 245 250 255
 Pro Met Asn Asp His Asn Thr Tyr Arg Cys Phe Ser Gln Pro Arg His
 30 260 265 270
 Ile Ser Val Ala Met Asp Lys Phe Gly Phe Ser Leu Pro Tyr Val Gln
 275 280 285
 Tyr Phe Gly Gly Val Ser Ala Leu Ser Lys Gln Gln Phe Leu Ser Ile
 290 295 300
 35 Asn Gly Phe Pro Asn Asn Tyr Trp Gly Trp Gly Gly Glu Asp Asp Asp
 305 310 315 320
 Ile Tyr Asn Arg Leu Ala Phe Arg Gly Met Ser Val Ser Arg Pro Asn
 325 330 335
 Ala Val Ile Gly Lys Cys Arg Met Ile Arg His Ser Arg Asp Lys Lys
 40 340 345 350
 Asn Glu Pro Asn Pro Gln Arg Phe Asp Arg Ile Ala His Thr Lys Glu
 355 360 365

Thr Met Leu Ser Asp Gly Leu Asn Ser Leu Thr Tyr Met Val Leu Glu
370 375 380
Val Gln Arg Tyr Pro Leu Tyr Thr Lys Ile Thr Val Asp Ile Gly Thr
385 390 395 400
5Pro Ser

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<213> Homo sapiens

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Gln Gly Gly Ser Asn Ser Ala Ala Ala Ile Gly Gln Ser Ser Gly Asp
20 25 30
Leu Arg Thr Gly Gly Ala Arg Pro Pro Pro Pro Leu Gly Ala Ser Ser
35 40 45
20Gln Pro Arg Pro Gly Gly Asp Ser Ser Pro Val Val Asp Ser Gly Pro
50 55 60
Gly Pro Ala Ser Asn Leu Thr Ser Val Pro Val Pro His Thr Thr Ala
65 70 75 80
Leu Ser Leu Pro Ala Cys Pro Glu Glu Ser Pro Leu Leu Val Gly Pro
25 85 90 95
Met Leu Ile Glu Phe Asn Met Pro Val Asp Leu Glu Leu Val Ala Lys
100 105 110
Gln

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<211> 85
<212> PRT
<213> Bos taurus
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Gln Gly Ser Ser His Gly Ala Ala Ala Ile Gly Gln Pro Ser Gly Glu
40 20 25 30
Leu Arg Leu Arg Gly Val Ala Pro Pro Pro Leu Gln Asn Ser Ser
35 40 45

Lys Pro Arg Ser Arg Ala Pro Ser Asn Leu Asp Ala Tyr Ser His Pro
50 55 60
Gly Pro Gly Pro Gly Pro Gly Ser Asn Leu Thr Ser Ala Pro Val Pro
65 70 75 80
5Ser Thr Thr Thr Arg
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<210> 9
<211> 273
10<212> PRT
<213> Homo sapiens

<400> 9
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Leu Ile Glu Phe Asn Met Pro Val Asp Leu Glu Leu Val Ala Lys Gln
20 25 30
Asn Pro Asn Val Lys Met Gly Gly Arg Tyr Ala Pro Arg Asp Cys Val
35 40 45
20Ser Pro His Lys Val Ala Ile Ile Pro Phe Arg Asn Arg Gln Glu
50 55 60
His Leu Lys Tyr Trp Leu Tyr Tyr Leu His Pro Val Leu Gln Arg Gln
65 70 75 80
Gln Leu Asp Tyr Gly Ile Tyr Val Ile Asn Gln Ala Gly Asp Thr Ile
25 85 90 95
Phe Asn Arg Ala Lys Leu Leu Asn Val Gly Phe Gln Glu Ala Leu Lys
100 105 110
Asp Tyr Asp Tyr Thr Cys Phe Val Phe Ser Asp Val Asp Leu Ile Pro
115 120 125
30Met Asn Asp His Asn Ala Tyr Arg Cys Phe Ser Gln Pro Arg His Ile
130 135 140
Ser Val Ala Met Asp Lys Phe Gly Phe Ser Leu Pro Tyr Val Gln Tyr
145 150 155 160
Phe Gly Gly Val Ser Ala Ser Ser Lys Gln Gln Phe Leu Thr Ile Asn
35 165 170 175
Gly Phe Pro Asn Asn Tyr Trp Gly Trp Gly Gly Glu Asp Asp Asp Ile
180 185 190
Phe Asn Arg Leu Val Phe Arg Gly Met Ser Ile Ser Arg Pro Asn Ala
195 200 205
40Val Val Gly Thr Cys Arg Met Ile Arg His Ser Arg Asp Lys Lys Asn
210 215 220

Glu Pro Asn Pro Gln Arg Phe Asp Arg Ile Ala His Thr Lys Glu Thr
 225 230 235 240
 Met Leu Ser Asp Gly Leu Asn Ser Leu Thr Tyr Gln Val Leu Asp Val
 245 250 255
 5Gln Arg Tyr Pro Leu Tyr Thr Gln Ile Thr Val Asp Ile Gly Thr Pro
 260 265 270
 Ser

10<210> 10
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 <212> PRT
 <213> Bos taurus

15<400> 10
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 20Asn Pro Lys Val Lys Leu Gly Gly Arg Tyr Thr Pro Met Asp Cys Ile
 35 40 45
 Ser Pro His Lys Val Ala Ile Ile Ile Leu Phe Arg Asn Arg Gln Glu
 50 55 60
 His Leu Lys Tyr Trp Leu Tyr Tyr Leu His Pro Met Val Gln Arg Gln
 2565 70 75 80
 Gln Leu Asp Tyr Gly Ile Tyr Val Ile Asn Gln Ala Gly Glu Ser Met
 85 90 95
 Phe Asn Arg Ala Lys Leu Leu Asn Val Gly Phe Lys Glu Ala Leu Lys
 100 105 110
 30Asp Tyr Asp Tyr Asn Cys Phe Val Phe Ser Asp Val Asp Leu Ile Pro
 115 120 125
 Met Asn Asp His Asn Thr Tyr Arg Cys Phe Ser Gln Pro Arg His Ile
 130 135 140
 Ser Val Ala Met Asp Lys Phe Gly Phe Ser Leu Pro Tyr Val Gln Tyr
 35145 150 155 160
 Phe Gly Gly Val Ser Ala Leu Ser Lys Gln Gln Phe Leu Ser Ile Asn
 165 170 175
 Gly Phe Pro Asn Asn Tyr Trp Gly Trp Gly Gly Glu Asp Asp Asp Ile
 180 185 190
 40Tyr Asn Arg Leu Ala Phe Arg Gly Met Ser Val Ser Arg Pro Asn Ala
 195 200 205

Val Ile Gly Lys Cys Arg Met Ile Arg His Ser Arg Asp Lys Lys Asn
210 215 220
Glu Pro Asn Pro Gln Arg Phe Asp Arg Ile Ala His Thr Lys Glu Thr
225 230 235 240
5Met Leu Ser Asp Gly Leu Asn Ser Leu Thr Tyr Met Val Leu Glu Val
245 250 255
Gln Arg Tyr Pro Leu Tyr Thr Lys Ile Thr Val Asp Ile Gly Thr Pro
260 265 270

Ser

10

<210> 11

<211> 1197

<212> PRT

15<213> Homo sapiens

<400> 11

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20 25 30
Cys Gly Cys Cys Gly Ala Thr Gly Cys Cys Ala Gly Gly Cys
35 40 45
Gly Cys Gly Thr Cys Cys Cys Thr Ala Cys Ala Gly Cys Gly Gly
25 50 55 60
Cys Cys Thr Gly Cys Cys Cys Thr Gly Cys Thr Cys Gly Thr
65 70 75 80
Gly Gly Cys Cys Gly Thr Cys Thr Gly Cys Gly Cys Thr Cys Thr Gly
85 90 95
30Cys Ala Cys Cys Thr Thr Gly Gly Cys Gly Thr Cys Ala Cys Cys Cys
100 105 110
Thr Cys Gly Thr Thr Ala Cys Thr Ala Cys Cys Thr Gly Gly Cys
115 120 125
Thr Gly Gly Cys Cys Gly Ala Cys Cys Thr Gly Ala Gly Cys
35 130 135 140
Cys Gly Cys Cys Thr Gly Cys Cys Cys Ala Ala Cys Thr Gly Gly
145 150 155 160
Thr Cys Gly Gly Ala Gly Thr Cys Thr Cys Cys Ala Cys Ala Cys Cys
165 170 175
40Gly Cys Thr Gly Cys Ala Gly Gly Cys Gly Gly Thr Cys Gly
180 185 190

10

Ala Ala Cys Ala Gly Thr Gly Cys Cys Gly Cys Cys Gly Cys Ala
 195 200 205
 Thr Cys Gly Gly Cys Ala Gly Thr Cys Cys Thr Cys Cys Gly Gly
 210 215 220
 SGly Gly Ala Cys Cys Thr Cys Cys Gly Ala Cys Cys Gly Gly Ala
 225 230 235 240
 Gly Gly Gly Cys Cys Gly Cys Gly Cys Cys Gly Cys Cys Gly Cys
 245 250 255
 Cys Thr Cys Cys Thr Cys Thr Ala Gly Gly Cys Gly Cys Cys Thr Cys
 10 260 265 270
 Cys Thr Cys Cys Ala Gly Cys Cys Gly Cys Cys Cys Gly
 275 280 285
 Gly Gly Thr Gly Gly Cys Gly Ala Cys Thr Cys Cys Ala Gly Cys Cys
 290 295 300
 15Cys Ala Gly Thr Cys Gly Thr Gly Gly Ala Thr Thr Cys Thr Gly Gly
 305 310 315 320
 Cys Cys Cys Thr Gly Gly Cys Cys Cys Gly Cys Thr Ala Gly Cys
 325 330 335
 Ala Ala Cys Thr Thr Gly Ala Cys Cys Thr Cys Gly Gly Thr Cys Cys
 20 340 345 350
 Cys Ala Gly Thr Gly Cys Cys Cys Cys Ala Cys Ala Cys Cys Ala Cys
 355 360 365
 Cys Gly Cys Ala Cys Thr Gly Thr Cys Gly Cys Thr Gly Cys Cys Cys
 370 375 380
 25Gly Cys Cys Thr Gly Cys Cys Cys Thr Gly Ala Gly Gly Ala Gly Thr
 385 390 395 400
 Cys Cys Cys Cys Gly Cys Thr Gly Cys Thr Thr Gly Thr Gly Gly
 405 410 415
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 Thr Thr Thr Ala Ala Cys Ala Thr Gly Cys Cys Thr Gly Thr Gly Gly
 435 440 445
 Ala Cys Cys Thr Gly Gly Ala Gly Cys Thr Cys Gly Thr Gly Cys
 450 455 460
 35Ala Ala Ala Gly Cys Ala Gly Ala Ala Cys Cys Cys Ala Ala Ala Thr
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 Gly Thr Gly Ala Ala Gly Ala Thr Gly Gly Cys Gly Gly Cys Cys
 485 490 495
 Gly Cys Thr Ala Thr Gly Cys Cys Cys Cys Ala Gly Gly Ala
 40 500 505 510
 Cys Thr Gly Cys Gly Thr Cys Thr Cys Thr Cys Cys Thr Cys Ala Cys
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Ala Ala Gly Gly Thr Gly Gly Cys Cys Ala Thr Cys Ala Thr Cys Ala
 530 535 540

Thr Thr Cys Cys Ala Thr Thr Cys Cys Gly Cys Ala Ala Cys Cys Gly
 545 550 555 560

5Gly Cys Ala Gly Gly Ala Gly Cys Ala Cys Cys Thr Cys Ala Ala Gly
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Thr Ala Cys Thr Gly Gly Cys Thr Ala Thr Ala Thr Thr Ala Thr Thr
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Thr Gly Cys Ala Cys Cys Cys Ala Gly Thr Cys Cys Thr Gly Cys Ala
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Gly Cys Gly Cys Cys Ala Gly Cys Ala Gly Cys Thr Gly Gly Ala Cys
 610 615 620

Thr Ala Thr Gly Gly Cys Ala Thr Cys Thr Ala Thr Gly Thr Thr Ala
 625 630 635 640

15Thr Cys Ala Ala Cys Cys Ala Gly Gly Cys Gly Gly Ala Gly Ala
 645 650 655

Cys Ala Cys Thr Ala Thr Ala Thr Cys Ala Ala Thr Cys Gly Thr
 660 665 670

Gly Cys Thr Ala Ala Gly Cys Thr Cys Cys Thr Cys Ala Ala Thr Gly
 20 675 680 685

Thr Thr Gly Gly Cys Thr Thr Cys Ala Ala Gly Ala Ala Gly Cys
 690 695 700

Cys Thr Thr Gly Ala Ala Gly Gly Ala Cys Thr Ala Thr Gly Ala Cys
 705 710 715 720

25Thr Ala Cys Ala Cys Cys Thr Gly Cys Thr Thr Gly Thr Gly Thr
 725 730 735

Thr Thr Ala Gly Thr Gly Ala Cys Gly Thr Gly Gly Ala Cys Cys Thr
 740 745 750

Cys Ala Thr Thr Cys Cys Ala Ala Thr Gly Ala Ala Thr Gly Ala Thr
 30 755 760 765

Cys Ala Thr Ala Ala Thr Gly Cys Gly Thr Ala Cys Ala Gly Gly Thr
 770 775 780

Gly Thr Thr Thr Thr Cys Ala Cys Ala Gly Cys Cys Ala Cys Gly
 785 790 795 800

35Gly Cys Ala Cys Ala Thr Thr Cys Cys Gly Thr Thr Gly Cys Ala
 805 810 815

Ala Thr Gly Gly Ala Thr Ala Ala Gly Thr Thr Gly Gly Ala Thr
 820 825 830

Thr Cys Ala Gly Cys Cys Thr Ala Cys Cys Thr Thr Ala Thr Gly Thr
 40 835 840 845

Thr Cys Ala Gly Thr Ala Thr Thr Thr Gly Gly Ala Gly Gly Thr
 850 855 860

12

Gly Thr Cys Thr Cys Thr Gly Cys Thr Thr Cys Ala Ala Gly Thr Ala
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 Ala Ala Cys Ala Ala Cys Ala Gly Thr Thr Thr Cys Thr Ala Ala Cys
 885 890 895
 5Cys Ala Thr Cys Ala Ala Thr Gly Gly Ala Thr Thr Thr Cys Cys Thr
 900 905 910
 Ala Ala Thr Ala Ala Thr Thr Ala Thr Thr Gly Gly Gly Cys Thr
 915 920 925
 Gly Gly Gly Ala Gly Gly Ala Gly Ala Ala Gly Ala Thr Gly Ala
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 Thr Gly Ala Cys Ala Thr Thr Thr Thr Ala Ala Cys Ala Gly Ala
 945 950 955 960
 Thr Thr Ala Gly Thr Thr Thr Thr Ala Gly Gly Cys Ala
 965 970 975
 15Thr Gly Thr Cys Thr Ala Thr Ala Thr Cys Thr Cys Gly Cys Cys Cys
 980 985 990
 Ala Ala Ala Thr Gly Cys Thr Gly Thr Gly Gly Thr Cys Gly Gly Gly
 995 1000 1005
 Ala Cys Gly Thr Gly Thr Cys Gly Cys Ala Thr Gly Ala Thr Cys Cys
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 Gly Cys Cys Ala Cys Thr Cys Ala Ala Gly Ala Gly Ala Cys Ala Ala
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 1045 1050 1055
 25Cys Cys Thr Cys Ala Gly Ala Gly Gly Thr Thr Thr Gly Ala Cys Cys
 1060 1065 1070
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 30 1090 1095 1100
 Gly Ala Thr Gly Gly Thr Thr Gly Ala Ala Cys Thr Cys Ala Cys
 1105 1110 1115 1120
 Thr Cys Ala Cys Cys Thr Ala Cys Cys Ala Gly Gly Thr Gly Cys Thr
 1125 1130 1135
 35Gly Gly Ala Thr Gly Thr Ala Cys Ala Gly Ala Gly Ala Thr Ala Cys
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 Cys Cys Ala Thr Thr Gly Thr Ala Thr Ala Cys Cys Cys Ala Ala Ala
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<212> DNA
<213> Artificial Sequence

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<220>
<223> A synthetic primer

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10atcgggaaga cgcgtaacat ccggccactcg agagac 36

<210> 13
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<212> DNA
15<213> Artificial Sequence

<220>
<223> A synthetic primer

20<400> 13

atcgggaaga cgcggtgagat ccggccactcg agagac

36